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## RESEARCH ARTICLE

## Clinical Assessment of Superficial Digital Flexor Tendon (SDFT) Core Lesion Treated with Platelet Rich Plasma (PRP) in Donkeys (*Equus Asinus*)

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### Abstract

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Little reviews are available about detailed clinical scoring as a tool for assessment of tendonitis in equine. The present investigation was designed to evaluate the clinical assessment scores of pain, heat, swelling and lameness of induced SDFT core lesion in donkeys treated with intralesional injection of PRP. The present study was carried out on two groups (group-I) 12 donkeys treated by intratendinous injection of freshly prepared platelet rich plasma (PRP) and (group-II) 7 donkeys control group and treated by intratendinous injection of normal saline (Placebo). The Clinical assessment scores were performed before induction, 5<sup>th</sup>, 15<sup>th</sup>, 30<sup>th</sup>, 45<sup>th</sup>, 60<sup>th</sup> and 90<sup>th</sup> days post induction of core lesion. The clinical assessment of the PRP treated group revealed significant ( $p \leq 0.05$ ) improvement of local heat and grade of lameness at 15<sup>th</sup>, 30<sup>th</sup> and 45<sup>th</sup> days post induction in addition to significant ( $p \leq 0.05$ ) improvement of the pain reaction, midmetacarpal circumference (MM-C) and medio-lateral width (MM-MLW) at 15<sup>th</sup>, 30<sup>th</sup>, 45<sup>th</sup>, 60<sup>th</sup> and 90<sup>th</sup> days post induction; and significant ( $p \leq 0.05$ ) decreased of midmetacarpal dorso-palmar thickness (MM-DPTH) at 30<sup>th</sup> and 90<sup>th</sup> days post induction. The control group exhibited significant ( $p \leq 0.05$ ) decreased of the local heat at 45<sup>th</sup> and 60<sup>th</sup>; lameness at 45<sup>th</sup> and 90<sup>th</sup> days and pain reaction, MM-C, MM-MLW and MM-DPTH at 90<sup>th</sup> days post induction. Clinical assessment scores of pain, heat, swelling and lameness in the present investigations proved that these parameters could be valuable in the assessment of SDFT core lesions and monitoring the treatment of tendopathy in equines.

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## INTRODUCTION

The superficial digital flexor tendinitis is a problem of a huge concern with high incidence among the musculoskeletal injuries in equine and represent about 8-43% (Vergari, et al., 2012 and Bazzano, et al., 2013). About 43-93% of affected horses sustaining reinjury and get early retirement and only about 20-60% of the affected horses were returned successfully to racing after long period of layoff and rehabilitation lasts from 6 up to 18 months. The inescapable subsequent result is harsh economic losses in equine industry (Carvalho et al., 2013 and Zuffova, et al., 2013).

Careful clinical evaluation of tendinitis is of significant value as the first and stress-free step for diagnosis and follows up of tendinitis (**Muttini et al., 2013**). The typical clinical feature of tendinitis characterized by local swelling, increased local heat, painful reaction on digital palpation and variable degree of lameness (**Bosch, 2010**).

Heat and pain associated with tendonitis could be assessed according to 4-point scale (**Schramme, et al., 2010**) from normal to severe (normal, 0; mild, 1; moderate, 2 and severe, 3). Accurate assessment of the mid-metacarpal swelling was achieved through measuring of the mid-metacarpal circumferences, medio-lateral width and the dorso-palmar thickness (**Menarima, et al., 2012**). Lameness grade was assessed by (**AAEP Guide, 1999**) to be from 0 to 5 according to its severity.

The regenerative therapy in form of PRP was get the way to fulfill the regenerative mission via their own growth factors which released just after injection of the PRP into the injured tendon (**Zandim, et al., 2013**). The intralesional use of PRP for treatment of SDFT tendinitis showed rapid healing and tremendous improvement over other current techniques of treatment (**Bazzano, et al., 2013**). The aim of the present study was to create a core lesion within the mid SDFT; monitoring the clinical assessment of grade of lameness, swelling, hotness score and pain reaction score after treatment with Platelet rich plasma (PRP).

## Materials and Methods

### *Animals*

Nineteen clinically normal adult donkeys (*Equus Asinus*) with the mean age  $5.9 \pm 0.38$  years and the mean weight  $135.45 \pm 2.2$  kg were included in this study. All animals were evaluated clinically and underwent bilateral ultrasonography examination of the SDFT to exclude tendon affections. The present study was approved by the Committee of Animal Welfare and Ethics, Faculty of Veterinary Medicine, Benha University.

Donkeys were randomly allocated into two groups:

**Group-I:** Consisted of 12 donkeys; core lesions were induced in the mid SDFT of right forelimb and received treatment with intralesional injection of freshly prepared PRP at 5<sup>th</sup>; 15<sup>th</sup> and 30<sup>th</sup> days post induction.

**Group-II:** Consisted of 7 animals; core lesions were induced in the mid SDFT of right forelimb and received treatment with intralesional injection of Placebo (normal saline) as control group.

### *Creation of SDFT Core lesion*

Surgery was performed under general anesthesia and complete aseptic condition according to **Schramme, et al., 2010** (Fig. 1). A small (2cm) horizontal skin incision was performed at the palmar aspect of the metacarpal bone 4 cm above the proximal sesamoid bones. Under ultrasonography guidance, a 2 mm blunt Kershner wire adjusted to a bone drill was used to create a tunnel over a distance of 3 cm parallel to the direction of the SDFT fiber. Thereafter, dental carbide burr (2 mm) adjusted to the bone drill was introduced along the previously created tunnel and moved forward and backward for three times until a consistent core defect was identified ultrasonographically (Fig. 1). Finally, the subcutaneous tissue and skin were routinely closed, the operated limbs were bandaged and the animals were left for 5 days in the resting box until the treatment protocols were started.

### *Preparation and Injection of Platelet Rich Plasma (PRP)*

Under complete aseptic condition, blood was collected directly from the jugular vein on sodium citrate anticoagulant in a ratio (6 ml sodium citrate for 34 ml blood) by using of 18 gauge needle adjusted to sterile syringe. Thereafter, the PRP was prepared according to technique described by **Bosch et al, 2009**. All steps of centrifugation (the first at 4800 rpm/20 min. 4c° and the second at 2400 rpm/15 min. 4c°) were achieved by using of swinging cooling digital centrifuge (*Sigma Laborzentrifugen GmbH, Germany*), all steps of manual separation was performed under the laminar airflow device (*Clindia, Belgium*) to provides a sterile condition for all steps of preparation and the total platelets count was determined in whole blood and the PRP by using of automatic cell counter system (*Esco Micro Pte. Ltd., Changi, Singapore*).

Just prior to the injection, activation of PRP was achieved by addition of calcium chloride 10% in ratio (0.1 ml calcium chloride to 1ml PRP). Sedation and high 4 point block was achieved; PRP was rapidly injected intralesionally by 18 gauge needle under ultrasonographic guidance (Fig. 2) till complete filling of the core defect (3-4 ml).

### *Clinical examination*

The clinical points of interest were evaluated at 5<sup>th</sup>; 15<sup>th</sup>; 30<sup>th</sup>; 45<sup>th</sup>; 60<sup>th</sup> and 90<sup>th</sup> days post induction and recorded in clinical score index (Table, 1).

**Table (1): Clinical Examination Score Index (Schramme, et al., 2010).**

Parameters, Method of Examination & Score	Before Induction	5 <sup>th</sup>	15 <sup>th</sup>	30 <sup>th</sup>	45 <sup>th</sup>	60 <sup>th</sup>	90 <sup>th</sup>
		day	day	day	day	day	day
<b>Animal Posture</b> (Inspection)	---	---	---	---	---	---	---
<b>MM-C</b> (Plastic Measuring Tape)	---	---	---	---	---	---	---
<b>MM- DPTH</b> (Hoof Caliber)	---	---	---	---	---	---	---
<b>MM-MLW</b> (Hoof Caliber)	---	---	---	---	---	---	---
<b>Tendon Hotness</b> (Digital Palpation & Thermal band) (Score 0 to 3) or (Normal, 36°; Mild, 37°; Moderate, 38° or Severe, 39°)	---	---	---	---	---	---	---
<b>Tendon Pain Reaction</b> (Digital Pressure) (Score 0 to 3) or (Normal, Mild, Moderate or Severe)	---	---	---	---	---	---	---
<b>Lameness</b> (Inspection) (Grade 1 to 5)	---	---	---	---	---	---	---

### Statistical analysis

Data was statistically analyzed by One-Way ANOVA with post-hock Duncan multiple comparison test using statistical software program (*SPSS for windows version 20, USA*). Differences were considered significant at ( $p \leq 0.05$ ).

### Results

The mean platelet count in the whole blood was  $148.17 \pm 15.8 \times 10^6$  platelets / ml. The mean platelet count in the PRP samples was  $437.0 \pm 31.8 \times 10^6$  platelets / ml. Platelet counts were ranged from 2.1 to 5.9 with the mean  $3.4 \pm 0.4$  fold greater in PRP compared to whole blood.

Clinical examinations conducted after induction of the mid SDFT core lesion revealed a clear evidence of increased local temperature, pain reaction on digital pressure, swelling of the metacarpal region and lameness.

#### Local Heat (Increased Local Temperature)

Physical examination were conducted at the 5<sup>th</sup> day post induction of the core lesion revealed that, the local temperature was severely increased to its maximum value (score 3) in both groups (Table, 2).

The PRP treated group (Table, 2) displayed mild to moderate degree of local heat during 15<sup>th</sup> and 30<sup>th</sup> days post induction with the mean score  $1.75 \pm 0.13$  and  $1.11 \pm 0.11$  respectively. During the 45<sup>th</sup> day, absence of local heat (score 0) was rapidly observed in four animals and the other two animals showed mild degree (score 1) with the mean score ( $0.33 \pm 0.21$ ). The local heat completely resolved at the 60<sup>th</sup> and 90<sup>th</sup> days post induction.

The control group (Table, 2) displayed moderate to severe degree of local heat at 15<sup>th</sup> and 30<sup>th</sup> days and mild to moderate degree of local heat at 45<sup>th</sup> day. During the 60<sup>th</sup> day post induction, absence of the local heat (score 0) was observed in two animals and the other two animals showed mild degree (score 1). The local heat completely resolved at the 90<sup>th</sup> day post induction.

The PRP treated group was significant ( $p \leq 0.05$ ) decreased at 15<sup>th</sup>, 30<sup>th</sup> and 45<sup>th</sup> days. Th control group showed significant ( $p \leq 0.05$ ) decreased at 45<sup>th</sup> and 60<sup>th</sup> days. Comparison between the PRP treated and control groups revealed that, the PRP treated group was significant ( $p \leq 0.05$ ) lower than the control group at 15<sup>th</sup>, 30<sup>th</sup>, 45<sup>th</sup> and 60<sup>th</sup> days post induction.

**Table (2):** Mean  $\pm$  Standard Error (M  $\pm$  SE) of the local heat scores for both groups at different time points after induction of the core lesion.

Time	Control Group (n=7)	PRP Treated Group (n=12)
5 days	3.00 $\pm$ 0.00 <sup>1 c</sup>	3.00 $\pm$ 0.00 <sup>1 d</sup>
15 days	2.83 $\pm$ 0.17 <sup>2 c</sup>	1.75 $\pm$ 0.13 <sup>1 c</sup>
30 days	2.40 $\pm$ 0.24 <sup>2 c</sup>	1.11 $\pm$ 0.11 <sup>1 b</sup>
45 days	1.50 $\pm$ 0.29 <sup>2 b</sup>	0.33 $\pm$ 0.21 <sup>1 a</sup>
60 days	0.50 $\pm$ 0.29 <sup>2 a</sup>	0.00 $\pm$ 0.00 <sup>1 a</sup>
90 days	0.00 $\pm$ 0.00 <sup>1 a</sup>	0.00 $\pm$ 0.00 <sup>1 a</sup>

Means of different parameter for the different groups within the same column or within the same row having different superscripts (letters or numbers respectively) are significantly different at level ( $p \leq 0.05$ ).

### *Pain Reaction on Digital Pressure*

Physical examination were conducted during the 5<sup>th</sup> day post induction of the core lesion revealed that, the pain reaction on digital pressure was severely increased to its maximum value (score 3) in both groups (Table, 3).

The PRP treated group (Table, 3) exhibited moderate to severe degree of pain reaction during the 15<sup>th</sup> day post induction with the mean pain score (2.42  $\pm$  0.15). During the 30<sup>th</sup> day post induction, mild to moderate degree of pain reaction was earlier aroused with the mean score (1.89  $\pm$  0.11). During the 45<sup>th</sup> day post induction, the pain reaction was mild degree (scored 1) in all animals. During the 60<sup>th</sup> day post induction, absence of the pain reaction (score 0) was observed in three animals and the other three animals was mild degree (score 1) with the mean score (0.5  $\pm$  0.22). The local pain reaction was completely resolved at 90<sup>th</sup> days post induction.

The control group (Table, 3) showed severe degree of pain reaction during the 5<sup>th</sup>, 15<sup>th</sup> and 30<sup>th</sup> days post induction. Moderate to severe degree of pain reaction was elicited during the 45<sup>th</sup> and 60<sup>th</sup> days post induction. During the 90<sup>th</sup> days, mild to moderate pain reaction was aroused.

The PRP treated group showed significantly ( $p \leq 0.05$ ) decreased at the 15<sup>th</sup>, 30<sup>th</sup>, 45<sup>th</sup>, 60<sup>th</sup> and 90<sup>th</sup> days. The control group showed significant ( $p \leq 0.05$ ) decreased at the 90<sup>th</sup> day. Comparison between the PRP treated and control groups revealed that, the PRP treated group was significant ( $p \leq 0.05$ ) lower than the control group during the 15<sup>th</sup>, 30<sup>th</sup>, 45<sup>th</sup>, 60<sup>th</sup> and 90<sup>th</sup> day post induction.

**Table (3):** Mean  $\pm$  Standard Error (M  $\pm$  SE) of the local pain reaction score for both groups at different time points after induction of the core lesion.

Time	Control Group (n=7)	PRP Treated Group (n=12)
5 days	3.00 $\pm$ 0.00 <sup>1 c</sup>	3.00 $\pm$ 0.00 <sup>1f</sup>
15 days	3.00 $\pm$ 0.00 <sup>2 c</sup>	2.42 $\pm$ 0.15 <sup>1 c</sup>
30 days	3.00 $\pm$ 0.00 <sup>2 c</sup>	1.89 $\pm$ 0.11 <sup>1d</sup>
45 days	2.75 $\pm$ 0.25 <sup>2 b c</sup>	1.00 $\pm$ 0.00 <sup>1 c</sup>
60 days	2.50 $\pm$ 0.29 <sup>2 b</sup>	0.50 $\pm$ 0.22 <sup>1b</sup>
90 days	1.33 $\pm$ 0.33 <sup>2 a</sup>	0.00 $\pm$ 0.00 <sup>1 a</sup>

Means of different parameter for the different groups within the same column or within the same row having different superscripts (letters or numbers respectively) are significantly different at level ( $p \leq 0.05$ ).

### *Lameness*

All animals were observed recumbent most of the time, exerted great efforts to stand and the animal's stances characterized by flexion of the carpal joint, and resting on the toe (Fig. 3). The PRP treated group showed these signs

during the first two weeks and in the control group signs were persisted for five weeks post induction. Then, the standing animals showed less weight bearing on the affected limb by kept it advanced forward (Fig. 3). During the 90<sup>th</sup> day post induction, all animals in PRP treated group were mostly seen in standing position in comparison with control group.

Lameness (Table, 4) during the 5<sup>th</sup> day post induction in PRP treated and control group displayed a pronounced degree of lameness (grade 3 to 4) with the mean grade of  $3.67 \pm 0.18$  and  $3.71 \pm 0.18$ , respectively.

Grade of lameness had gradually decreased in PRP treated group (Table, 4) from 2 to 3 (mean;  $2.33 \pm 0.14$ ) during the 15<sup>th</sup> day post induction and from 1 to 2 (mean;  $1.56 \pm 0.18$ ) during 30<sup>th</sup> day. During 45<sup>th</sup> day post induction, lameness was disappeared in one animal and the other five animals showed grade 1 (mean;  $0.83 \pm 0.17$ ). Lameness was completely disappeared during the 60<sup>th</sup> and 90<sup>th</sup> days post induction.

Grade of lameness (Table, 4) in control group was 3 to 4 during the 15<sup>th</sup> and 30<sup>th</sup> days post induction. During the 45<sup>th</sup> and 60<sup>th</sup> days post induction, lameness was graded from 2 to 3 with the mean grade  $2.33 \pm 0.21$  and  $2.17 \pm 0.17$ , respectively. During 90<sup>th</sup> day post induction, lameness was graded from 1 to 2 with mean grade ( $1.33 \pm 0.33$ ).

The PRP treated group showed significantly ( $p \leq 0.05$ ) decreased in grade of lameness at the 15<sup>th</sup>, 30<sup>th</sup> and 45<sup>th</sup> days. The control group showed significantly ( $p \leq 0.05$ ) decreased at the 45<sup>th</sup> and 90<sup>th</sup> days. Comparison between the PRP treated and control groups revealed that, the PRP treated group was significant ( $p \leq 0.05$ ) lower in grade of lameness than the control group at 15<sup>th</sup>, 30<sup>th</sup>, 45<sup>th</sup>, 60<sup>th</sup>, and 90<sup>th</sup> days post induction.

**Table (4):** Mean  $\pm$  Standard Error (M  $\pm$  SE) of lameness scores for both groups at different time points after induction of the core lesion.

	Control group (n=7)	PRP Treated Group (n=12)
<b>5 days</b>	$3.71 \pm 0.18$ <sup>1 c</sup>	$3.67 \pm 0.14$ <sup>1 c</sup>
<b>15 days</b>	$3.40 \pm 0.20$ <sup>2 c</sup>	$2.33 \pm 0.14$ <sup>1 d</sup>
<b>30 days</b>	$3.29 \pm 0.18$ <sup>2 c</sup>	$1.56 \pm 0.18$ <sup>1 c</sup>
<b>45 days</b>	$2.33 \pm 0.21$ <sup>2 b</sup>	$0.83 \pm 0.17$ <sup>1 b</sup>
<b>60 days</b>	$2.17 \pm 0.17$ <sup>2 b</sup>	$0.33 \pm 0.21$ <sup>a1 b</sup>
<b>90 days</b>	$1.33 \pm 0.33$ <sup>a2</sup>	$0.00 \pm 0.00$ <sup>a1</sup>

Means of different parameter for the different groups within the same column or within the same row having different superscripts (letters or numbers respectively) are significantly different at level ( $p \leq 0.05$ ).

### Swelling

The PRP treated and the control groups displayed diffuse swelling of the metacarpal region during the first 24hr hours post induction. During the 5<sup>th</sup> day post induction, the swelling was confined only to the palmar aspect of the midmetacarpal region giving the typical bow appearance “bowed tendon” when observed from the lateral view and wide fusiform appearance when viewed from the palmar aspect (Fig. 4).

This characteristic bow appearance was taken a wide posterior curve at all the length of the palmar aspect of the mid-metacarpal region and persisted in all animals of control group till the end of the study. During the 45<sup>th</sup> and 60<sup>th</sup> days post induction of the core lesion in PRP treated group, the characteristic bow appearance was subsided to be focal swelling as narrow posterior curve including the site of skin incision and narrow area proximal and distal (Fig. 4).

### Mid-Metacarpal Circumference

The mid metacarpal circumference (Table, 5) in the PRP treated group was severely increased from  $10.38 \pm 0.05$  cm before induction to  $13.18 \pm 0.09$  cm at the 5<sup>th</sup> day post induction. Thereafter it sharply decreased after the beginning of the treatment protocol to nearly the normal value ( $10.63 \pm 0.07$ ) at the 90<sup>th</sup> day post induction.

The mid metacarpal circumference in the control group (Table, 5) was severely increased from  $10.37 \pm 0.06$  cm before induction to its maximum value ( $13.25 \pm 0.09$  cm) at the 15<sup>th</sup> days post induction. Then it slightly decreased to  $12.27 \pm 0.15$  cm at 90<sup>th</sup> day post induction.

The mean mid metacarpal circumference in PRP treated group was significant ( $p \leq 0.05$ ) decreased at 15<sup>th</sup>, 30<sup>th</sup>, 45<sup>th</sup>, 60<sup>th</sup> and 90<sup>th</sup> days. The control group was significant ( $p \leq 0.05$ ) decreased at the 90<sup>th</sup> day post induction. Comparison between the PRP treated and the control groups revealed that, the PRP treated group was significantly ( $p \leq 0.05$ ) lower than the control during the 30<sup>th</sup>, 45<sup>th</sup>, 60<sup>th</sup> and 90<sup>th</sup> days post induction.

**Table (5):** Mean  $\pm$  Standard Error (M  $\pm$  SE) of mid-metacarpal circumference (cm) for both groups at different time points after induction of the core lesion.

Time	Control group (n=7)	PRP Treated Group (n=12)
Normal	$10.37 \pm 0.06^{a1}$	$10.38 \pm 0.05^{a1}$
5 days	$13.20 \pm 0.11^{1d}$	$13.18 \pm 0.09^{1f}$
15 days	$13.25 \pm 0.09^{2cd}$	$12.77 \pm 0.09^{1e}$
30 days	$13.00 \pm 0.07^{2cd}$	$12.30 \pm 0.11^{1d}$
45 days	$12.97 \pm 0.09^{2cd}$	$11.93 \pm 0.12^{1e}$
60 days	$12.83 \pm 0.09^{2c}$	$11.53 \pm 0.11^{1b}$
90 days	$12.27 \pm 0.15^{2b}$	$10.63 \pm 0.07^{a1}$

Means of different parameter for the different groups within the same column or within the same row having different superscripts (letters or numbers respectively) are significantly different at level ( $p \leq 0.05$ ).

#### *Mid-Metacarpal Medio-Lateral (width)*

The mean mid metacarpal medio-lateral width (Table, 6) in the PRP treated group was severely increased from  $1.41 \pm 0.04$  cm before induction to  $2.95 \pm 0.06$  cm at 5<sup>th</sup> day post induction. Then it sharply decreased after the beginning of the treatment protocol to nearly the normal value ( $1.5 \pm 0.15$ ) at the 90<sup>th</sup> day post induction.

The mean mid metacarpal medio-lateral width in the control group was severely increased from  $1.4 \pm 0.03$  cm before induction to its maximum value ( $2.98 \pm 0.02$  cm) at the 15<sup>th</sup> days post induction. Thereafter it slightly decreased to  $2 \pm 0.06$  cm at the 90<sup>th</sup> day post induction.

The mean mid metacarpal medio-lateral width in the PRP treated group was significant ( $p \leq 0.05$ ) decreased at 15<sup>th</sup>, 30<sup>th</sup>, 45<sup>th</sup>, 60<sup>th</sup> and 90<sup>th</sup> days. The control group was significant ( $p \leq 0.05$ ) decreased at 90<sup>th</sup> day. Comparison between the PRP treated and the control groups revealed that, the PRP treated group was significantly ( $p \leq 0.05$ ) lower than the control group at the 15<sup>th</sup>, 30<sup>th</sup>, 45<sup>th</sup>, 60<sup>th</sup> and 90<sup>th</sup> days post induction.

**Table (6):** Mean  $\pm$  Standard Error (M  $\pm$  SE) of mid-metacarpal medio-lateral width (cm) for both groups at different time points after induction of the core lesion.

Time	Control group (n=7)	PRP Treated Group (n=12)
Normal	$1.4 \pm 0.03^{a1}$	$1.41 \pm 0.04^{a1}$
5 days	$2.91 \pm 0.09^{1de}$	$2.95 \pm 0.06^{1f}$
15 days	$2.98 \pm 0.02^{2e}$	$2.73 \pm 0.06^{1e}$
30 days	$2.92 \pm 0.05^{2de}$	$2.42 \pm 0.08^{1d}$
45 days	$2.75 \pm 0.09^{2cd}$	$2.18 \pm 0.09^{1e}$
60 days	$2.65 \pm 0.12^{2c}$	$1.92 \pm 0.07^{1b}$

**90 days** $2.00 \pm 0.06^{2b}$  $1.50 \pm 0.15^{a1}$ 

Means of different parameter for the different groups within the same column or within the same row having different superscripts (letters or numbers respectively) are significantly different at level ( $p \leq 0.05$ ).

#### *Mid-Metacarpal Dorsopalmar (Thickness)*

The mean mid metacarpal dorsopalmar thickness (Table, 7) of the PRP treated group was severely increased from  $3.30 \pm 0.10$  cm before induction to  $5.08 \pm 0.05$  cm at 5<sup>th</sup> day post induction. Thereafter, it was sharply decreased after the beginning of the treatment protocol to nearly the normal value  $3.47 \pm 0.09$  at 90<sup>th</sup> day post induction.

The mid metacarpal dorsopalmar thickness of the control group (Table, 7) was severely increased from  $3.29 \pm 0.04$  cm before induction to its maximum value ( $5.07 \pm 0.07$  cm) at the 15<sup>th</sup> days post induction. Thereafter it slightly decreased to  $4.20 \pm 0.06$  cm at the 90<sup>th</sup> day post induction.

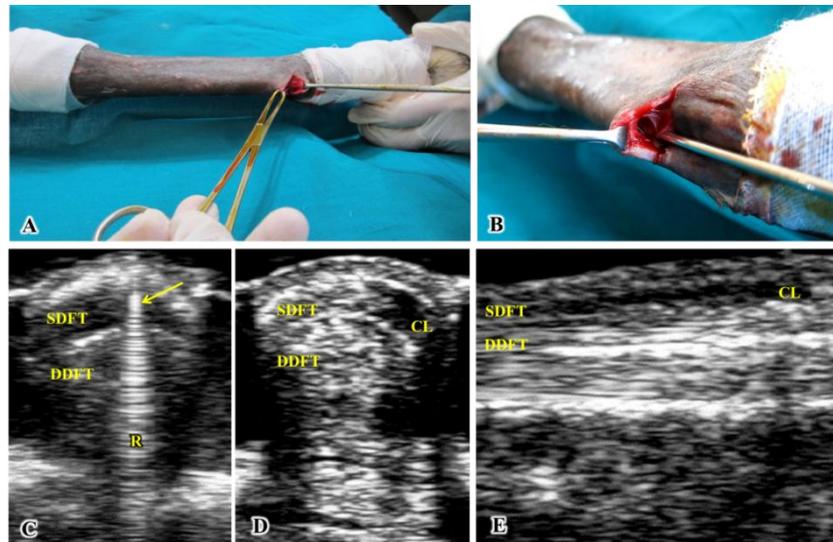
The mean mid metacarpal dorsopalmar thickness of the PRP treated group was significant ( $p \leq 0.05$ ) decreased at the 30<sup>th</sup> and 90<sup>th</sup> days. The control group was significant ( $p \leq 0.05$ ) decreased at 90<sup>th</sup> day. Comparison between the PRP treated and control groups revealed that, the PRP treated group was significant ( $p \leq 0.05$ ) lower than the control group at the 15<sup>th</sup>, 30<sup>th</sup>, 45<sup>th</sup>, 60<sup>th</sup> and 90<sup>th</sup> days post induction (Table, 7).

Statistical analysis of the mean mid metacarpal circumference, mean medio-lateral width and mean dorso-palmar thickness recorded at the 90<sup>th</sup> day in the PRP treated group showed a non-significant ( $p \leq 0.05$ ) difference in comparison with the mean normal value that recorded before induction. While both value in the control group were significant ( $p \leq 0.05$ ) different (Table, 5, 6 and 7).

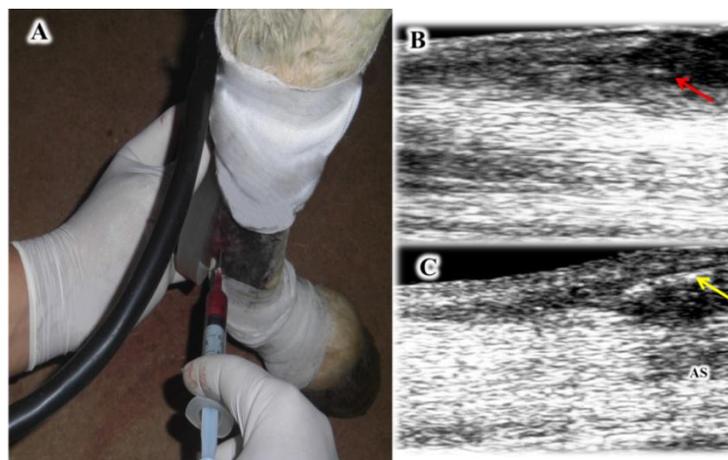
**Table (7):** Mean  $\pm$  Standard Error (M  $\pm$  SE) of mid-metacarpal dorso-palmar thickness (cm) for both groups at different time points after induction of the core lesion.

Time	Control group (n=7)	PRP Treated Group (n=12)
<b>Normal</b>	$3.29 \pm 0.04^{1a}$	$3.30 \pm 0.10^{1a}$
<b>5 days</b>	$5.06 \pm 0.09^{1c}$	$5.08 \pm 0.05^{1d}$
<b>15 days</b>	$5.07 \pm 0.07^{2c}$	$4.83 \pm 0.05^{1d}$
<b>30 days</b>	$5.00 \pm 0.11^{2c}$	$4.43 \pm 0.08^{1c}$
<b>45 days</b>	$4.90 \pm 0.07^{2c}$	$4.17 \pm 0.11^{1bc}$
<b>60 days</b>	$4.83 \pm 0.12^{2c}$	$3.93 \pm 0.11^{1b}$
<b>90 days</b>	$4.20 \pm 0.06^{2b}$	$3.47 \pm 0.09^{1a}$

Means of different parameter for the different groups within the same column or within the same row having different superscripts (letters or numbers respectively) are significantly different at level ( $p \leq 0.05$ ).



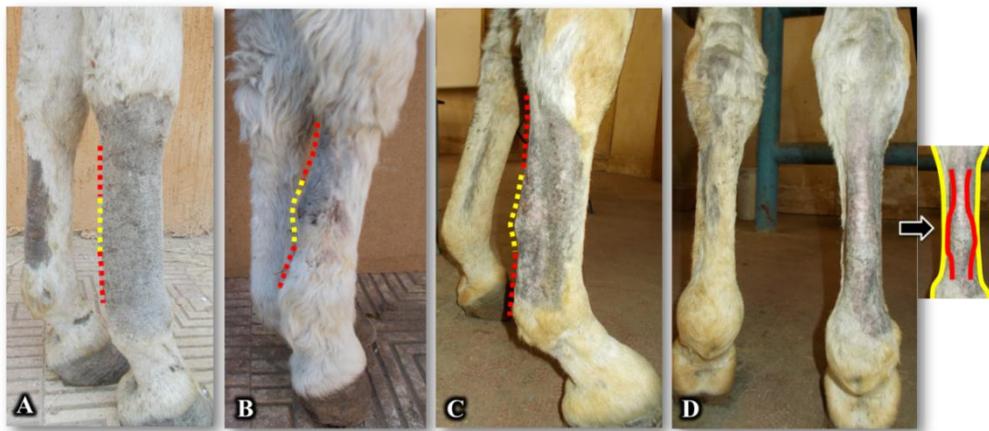
**Figure (1):** Surgical Procedure of Induction of Core Lesion in mid SDFT (A & B). Transverse ultrasonographic examination during induction (C), note the hyperechoic signal of the burr (yellow arrow) and the distal reverberation artifact (R). Transverse and sagittal ultrasonography after induction (D & E), note the anechoic core lesion (CL) after distention by normal saline.



**Figure (2):** Ultrasound-guided intralesional injection of PRP (A) into a core lesion of SDFT. Sagittal Plane of the SDFT core lesion before (B) and During (C) injection. Note, anechoic core lesion (red arrow) and hyper-echoic needle (yellow arrow) with distal acoustic shadowing (AS).



**Figure (3):** (A) Animal stance characterized by flexion of the carpal joint and resting on the toe. (B) Keeping the affected limb slight in front of the normal one.



**Figure (4):** Diffuse swelling of the metacarpal region (A). Typical bow appearance “bowed tendon” when viewed from the lateral view (B) and wide fusiform appearance of tendon when viewed from the palmar aspect (D). Focal swelling at the level of the mid metacarpal region (C).

## Discussion

In the present investigations clinical assessment of surgical induced mid SDFT core lesion revealed clear increased local temperature, pain reaction elicited on digital pressure, swelling of the metacarpal region and lameness. These findings coincided with (Marxen et al., 2004; Dahlgren, 2007; Bosch, 2010; De Lacerda Neto, et al., 2013 and Zandim 2013).

Smith and Goodship (2004) reported that to evaluate a tendon for an injury, clinicians need to assess the lameness, swelling and thickening, heat, response to digital palpation, and the appearance of tendon from multiple angles. In the present study clinical assessment and scoring of local heat, pain reaction, lameness and local swelling were conducted.

Accordingly, O’Sullivan (2007) and Fortier and Smith (2008) reported that, intratendinous PRP act to decrease the inflammatory process, enhancement tendon biological function, morphologically reconstitute the injured tissue by early granulation of defects and minimized scar tissue and adhesions. Furthermore, Georg, et al. (2010); Urrea-Chávez (2012); Zhang, et al. (2013); and Zuffova, et al. (2013) found that, growth factors released from the activated PRP included; platelet derived growth factor (PDGF), transforming growth factor (TGF),

fibroblast growth factor (FGF), vascular endothelial growth factor (VEGF), and hepatocyte growth factor (HGF) play an important role in cellular proliferation and differentiation within the injured tendinous tissues.

Therefore, the improvement in the scoring and parameters assessment in the present study could be attributed to the role of activated intratendinous injection of PRP. In addition to our findings in the scores assessment, **Zhang et al. (2013)** hypothesized that the hepatocyte growth factor (HGF) in PRP causes the anti-inflammatory effects which responsible for the pain, heat and swelling clearance effect of the PRP.

Lameness and pain in tendinitis was attributed to destruction of tendon fibers, pressure resulted from accumulation of blood and inflammatory exudates and high concentration of chemical irritants, like lactic acid (**Meimandi-Parizi et al., 2013**). Moreover, **Marxen et al., (2004)** attributed local swelling to the formation of intratendinous edema and influx of inflammatory cells, as a part of the inflammatory process.

Consequently, the induced core lesion assessment scores showed that the control group exhibited significant ( $p \leq 0.05$ ) decreased of the mean local heat at 45<sup>th</sup> and 60<sup>th</sup>; the mean lameness at 45<sup>th</sup> and 90<sup>th</sup> days; the mean pain reaction, MM-C, MM-MLW and MM-DPTH at 90<sup>th</sup> days post induction. Contrary to that, all the clinical assessment scores and parameters in the PRP treated group were significantly ( $p \leq 0.05$ ) lower than the control group at 15<sup>th</sup>, 30<sup>th</sup>, 45<sup>th</sup>, 60<sup>th</sup> and 90<sup>th</sup> days post induction.

Clinical assessment scores of pain, heat, swelling and lameness in the present investigations proved that these parameters could be useful in the assessment of SDFT core lesions during treatment of tendopathy in equines.

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